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DISTRIBUTION OF *EUGLENA OBTUSA* SCHMITZ AND *E. SALINA* LIEBETANZ ON THE AVON - HEATHCOTE ESTUARY, CHRISTCHURCH

D.A. STEFFENSEN

Botany Department, University of Canterbury, Christchurch, New Zealand

ABSTRACT

Aerial and ground surveys of the Avon-Heathcote Estuary indicated that euglenoid scum was concentrated near the source of effluent discharges, particularly the discharge from the Bromley oxidation ponds. Decreases in numbers of *Euglena* spp. in some regions of the Estuary since 1965 can be related to decreases in the amount of effluent discharged. Distribution up the Heathcote River was limited by a lack of suitable substrate rather than by low salinity, and water content of the mud appears to be less important than formerly suggested. The factor most consistently linked with numbers of *Euglena* was high carbon level in the mud. *E. obtusa* was rendered colourless by treatment with streptomycin, but remained active and at high concentrations in cultures for 3 months. This indicates it is probably heterotrophic, and may explain its occurrence on muds with high carbon content.

INTRODUCTION

Euglena was first recorded in the Avon-Heathcote Estuary by Linzey (1944), who noted "a thin yellow surface scum of some form of *Euglena* near the sewer outlets". The principal component of this scum is a light green, nonflagellated and "highly metabolic" (rapid changes in cell shape) species identified by Bruce (1953) as *Euglena limosa* Gard. This identification was based on morphological and habitat similarities with material described by Gard (1919) and Bracher (1919, 1929) from France and England respectively. Carter (1933) regarded the English material, previously named *E. dese* Ehrenberg by Brancher, as identical with *E. limosa*. Gojdics (1953), however, suggested *E. limosa* may be a synonym for *E. obtusa* Schmitz. This view is supported by G. Leedale (Palmer and Round 1965) who redescribed the English material as *E. obtusa* which is the name I propose to use for the Christchurch material. Williams (1960) recorded a second species, *E. salina* Liebetanz, from the Heathcote River. It is dark green and smaller than *E. obtusa*.

Descriptions of the extent of the scum and concentrations of cells have been included in the following biological surveys of the Estuary: Bruce (1953), Williams (1960), Rosenberg (1963), Webb (1965), Cameron (1968) and Marshall (1971). The present study was carried out in 1969 and 1972, the aim being to monitor any changes in patterns of distribution and to investigate factors affecting these patterns.

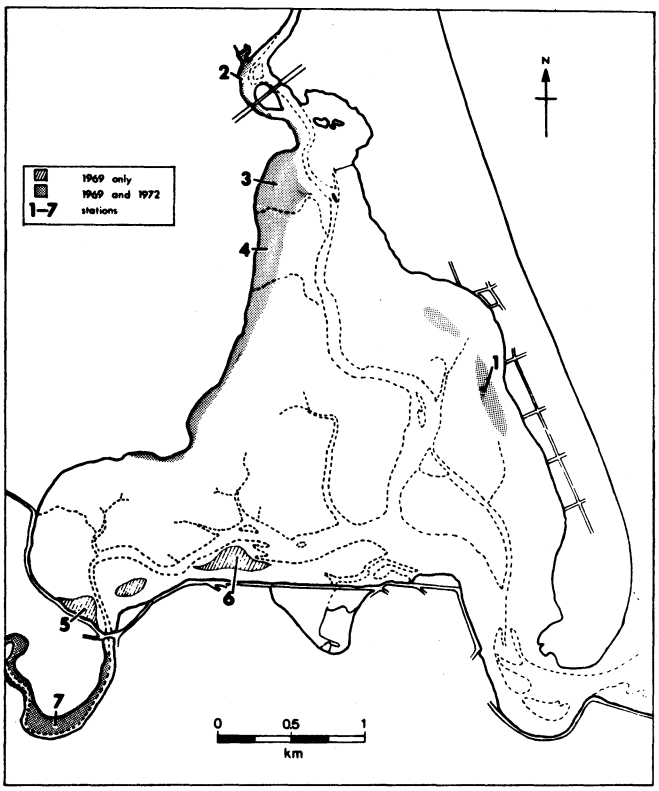

Fig. 1. *Euglena* distribution in the Avon-Heathcote Estuary in 1967 and 1972.

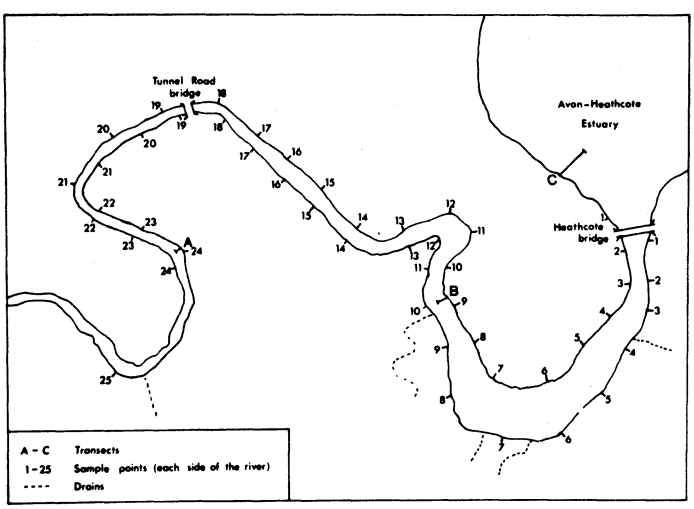

Fig. 2. Lower Heathcote River showing location of 49 sampling points and three transects.

METHODS

The area covered by scum was mapped from a ground survey in 1969 and from aerial colour photographs in 1972 (Fig. 1). Concentration of cells in the scum was sampled at seven stations (see Fig. 1). Numbers of cells were estimated from three surface samples collected on 100 mm^2 pieces of lens tissue at each station. A further three samples, 100 mm^2 and 3 mm deep, were also collected at each station. Preliminary work using frozen core samples had confirmed the observation of Palmer and Round (1965) that all *Euglena* was within 3 mm of the surface at low tide, the time when samples were taken. Each sample was diluted in 200 ml water and three 1 ml aliquots from each were counted in a plankton cell.

In 1969, a series of 24 sampling points were established at mid-tide level at 150 m intervals on each side of the Heathcote River to a point 3.6 km upstream from the Heathcote bridge (Fig. 2). A further sample point was established 4.2 m upstream from the bridge on the southern bank. These points correspond to the limits of firm mud banks. Three high- to low-tidal transects were established at Stations A, B, and C (see Fig. 2). At each sample point and transect level *Euglena* density was determined as above, and three 100 ml mud samples were taken and analysed for water and carbon content. Water content was estimated from weight loss after drying at 110°C for 48 hours. Carbon content was determined from weight loss after combustion at 600°C for 15 hours. Some samples were also analysed by the Schollenberg-Allison technique (Metson 1956) in which the sample is digested in concentrated H_2SO_4 and potassium dichromate, and titrated against ferrous ammonium sulphate.

Salinity of the overlying water was determined at low water neap with a salinometer.

Laboratory cultures were grown in water of different salinities ranging from seawater to freshwater at 18°C. Cell concentration was determined every second day for a period of two months.

RESULTS AND DISCUSSION

In 1969, scum formed a band along the high-tide mark on the western shore of the Estuary. An area of high concentration was centred on the outfall from the Bromley oxidation ponds and extended up the Avon and Heathcote Rivers. There were also thinner patches on the Brighton Spit and near Mt Pleasant. Densities ranged from 20 000 cells per 100 mm^2 on the Brighton Spit to over 250 000 cells per 100 mm^2 near the sewage outfalls. Approximately half the population was contained in the surface samples (Table 1).

The pattern in 1972 was similar to that found in previous surveys with high density patches occurring near the oxidation ponds. A lower concentration in the area adjacent to Mt Pleasant, St Andrews Hill, and west of the Heathcote River could be linked with removal of domestic sewer discharge from this area (in 1965 and 1966) and closing of a starch factory in 1968, which previously discharged highly organic effluent west of the Heathcote River. Over the rest of the Estuary the concentration had increased since 1953, but appears to have stabilised in

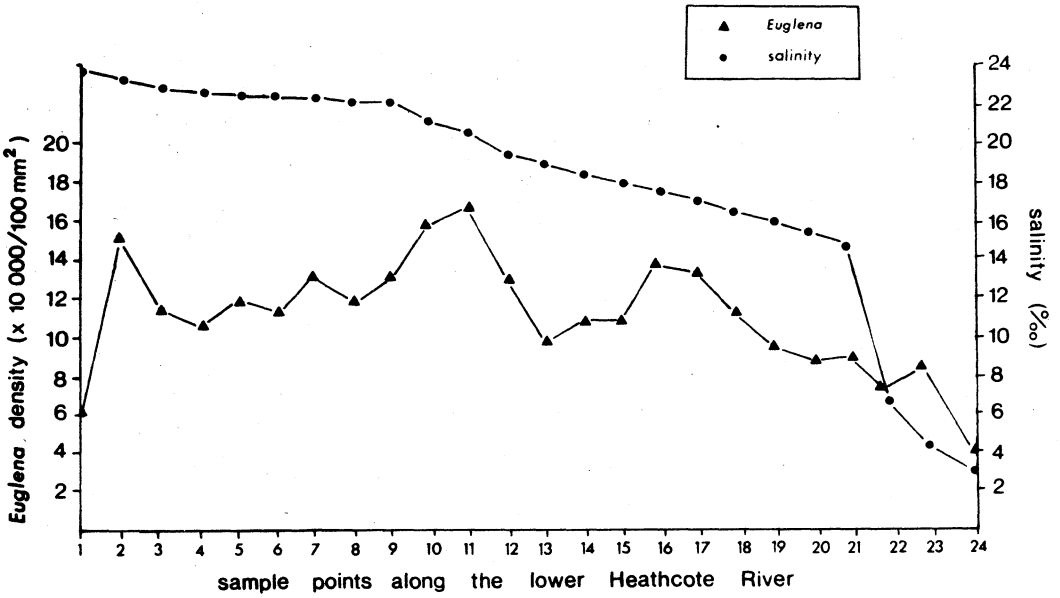

Fig. 3. *Euglena* density and salinity along the lower Heathcote River.

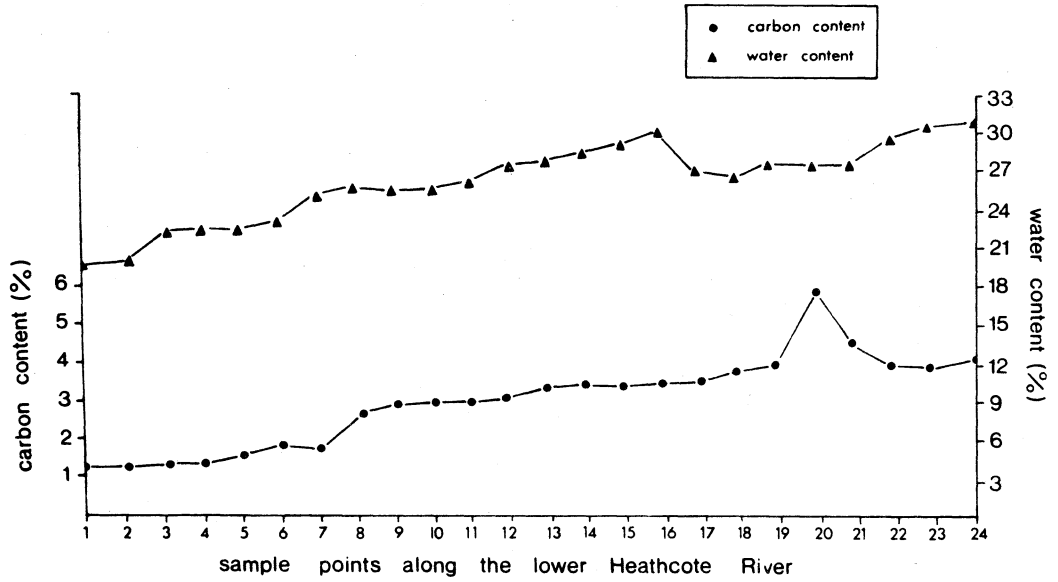

Fig. 4. Carbon and water content of substrate along the lower Heathcote River.

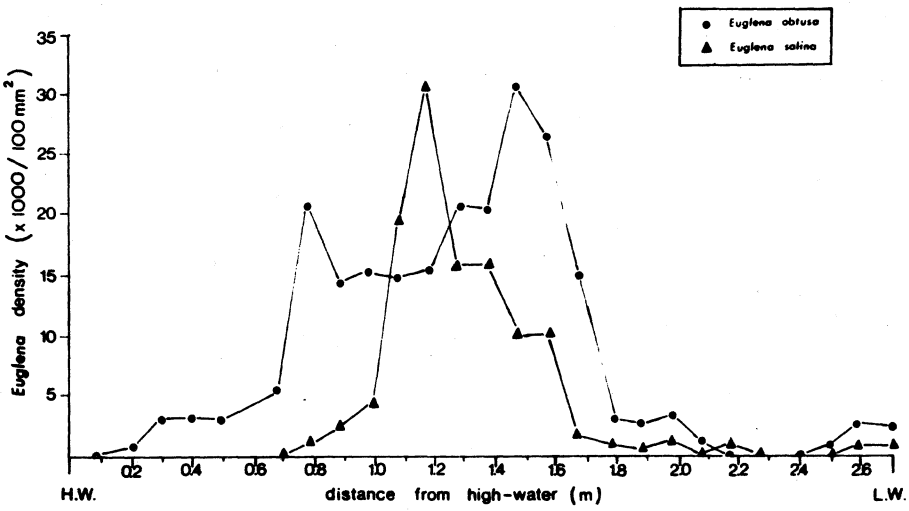

Fig. 5. Density of *Euglena obtusa* and *E. salina* along transect A.

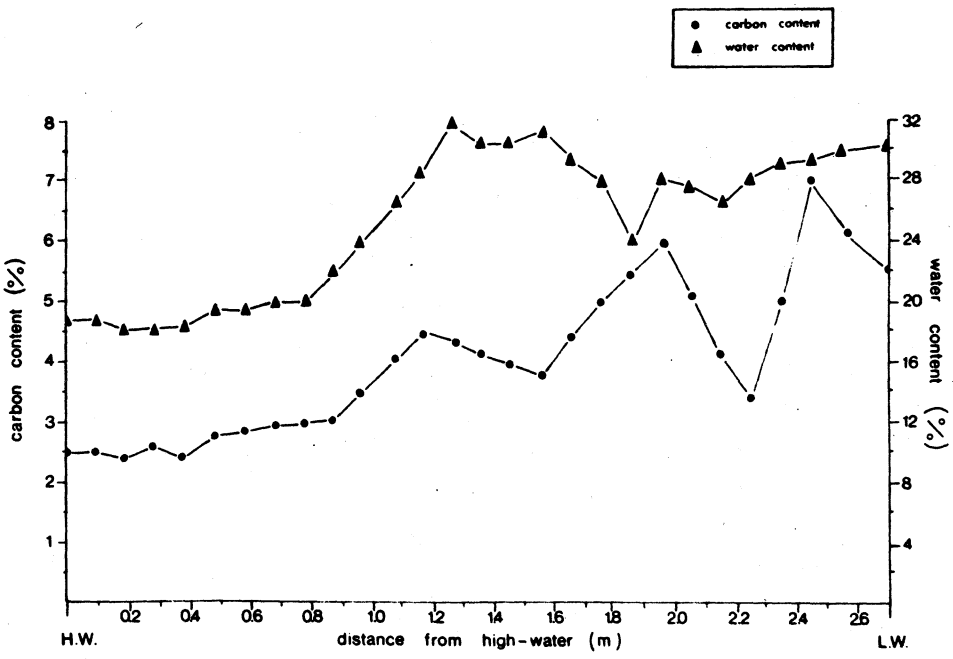

Fig. 6. Carbon and water content of substrate along transect A.

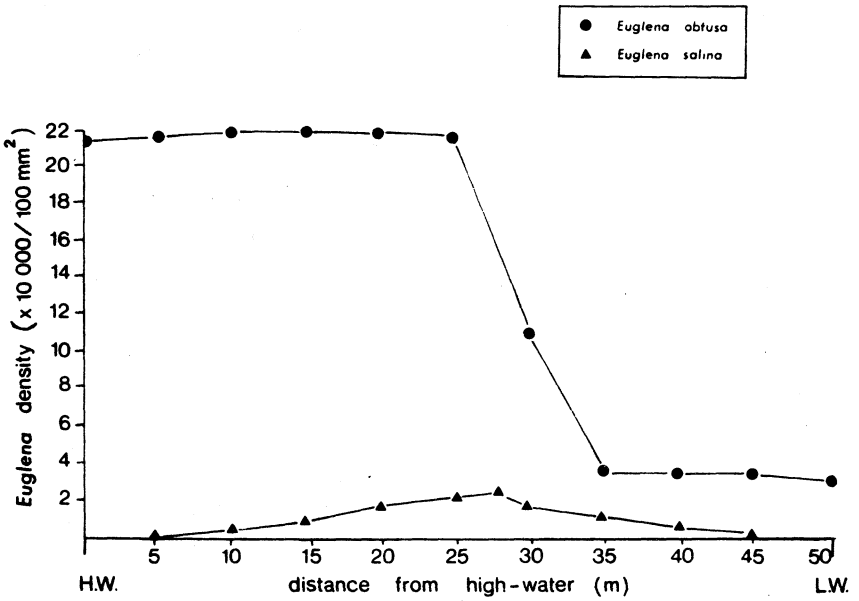

Fig. 7. Density of *Euglena obtusa* and *E. salina* along transect C.

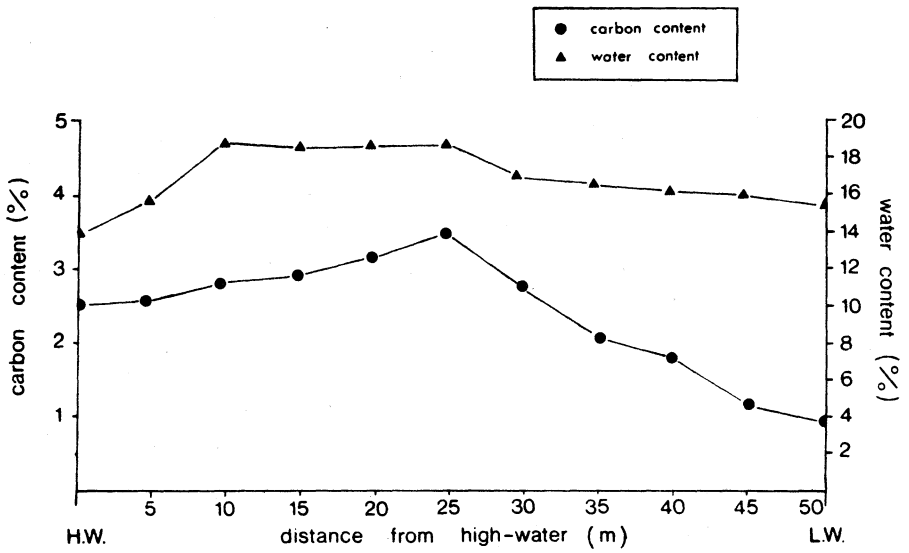

Fig. 8. Carbon and water content of substrate along transect C.

TABLE 1. *Euglena* per 100 m^2 in surface scum samples taken at seven stations in the Avon-Heathcote Estuary.

Station	Numbers at Surface (taken with lens tissue)	Total Numbers (to 3 mm in depth)
I	3 500	10 000
II	130 000	250 000
III	110 000	230 000
IV	153 000	260 000
V	130 000	220 000
VI	76 000	160 000
VII	11 000	20 000

All stations are significantly different from each other at 0.001 confidence level (Least Significant Difference = 7 922) except I and VII (L.S.D. = 6 742; significant at 0.05 confidence level) and II and V (no significant difference).

recent years.

Both species of *Euglena* occurred on the firm, almost flat intertidal mud bank which extends 3.6 km up the Heathcote River from the Heathcote Bridge. Numbers of *E. obtusa* at the sample points (Figs. 3, 4) tended to increase upstream from the Estuary with a maximum of 200 000 cells per 100 mm^2 at sample point 21. Above this point there was a rapid decline in numbers to 40 000 cells per 100 mm^2 at the furthest upstream extension of the bank. An isolated portion of flat intertidal bank a further 600 m upstream (sample point 25) had 23 000 cells per 100 mm^2. *E. salina* was present near the mouth of the river, and at upstream stations, but was absent from samples taken in the middle of the area sampled. Carbon and water content of the mud increased upstream along with *Euglena* numbers, but continued to increase after the latter declined. The decline in *Euglena* numbers corresponded with a sharp decline in salinity of the overlying water. Absence of *Euglena* between 3.6 km and 4.2 km from the Estuary corresponded to the limits of the flat intertidal mud bank.

Along transects A and B highest cell numbers occurred in the mid-tidal region of the mud bank, but numbers decreased rapidly towards high and low watermarks. The decreases at low watermark corresponded to the lower limit of firm mud. Below this was black, highly organic, almost liquid mud which apparently contained very little life of any description. The decline in *Euglena* numbers towards high-tide mark was associated with a sharp decline in carbon and water content (Figs. 5, 6).

Along transect C numbers of *E. obtusa* were very high near high-tide mark (over 200 000 cells per 100 mm^2), but decreased towards low-tide mark. Water and carbon content followed a similar pattern, but substrate remained firm along the full length of the transect (Figs 7, 8). Salinity was 24‰ at all points.

In laboratory cultures *E. obtusa* survived in salinities from 1‰ to 36‰ but higher concentrations were maintained in the 10 to 25‰ salinity range (Fig. 9). *E. salina* did not survive

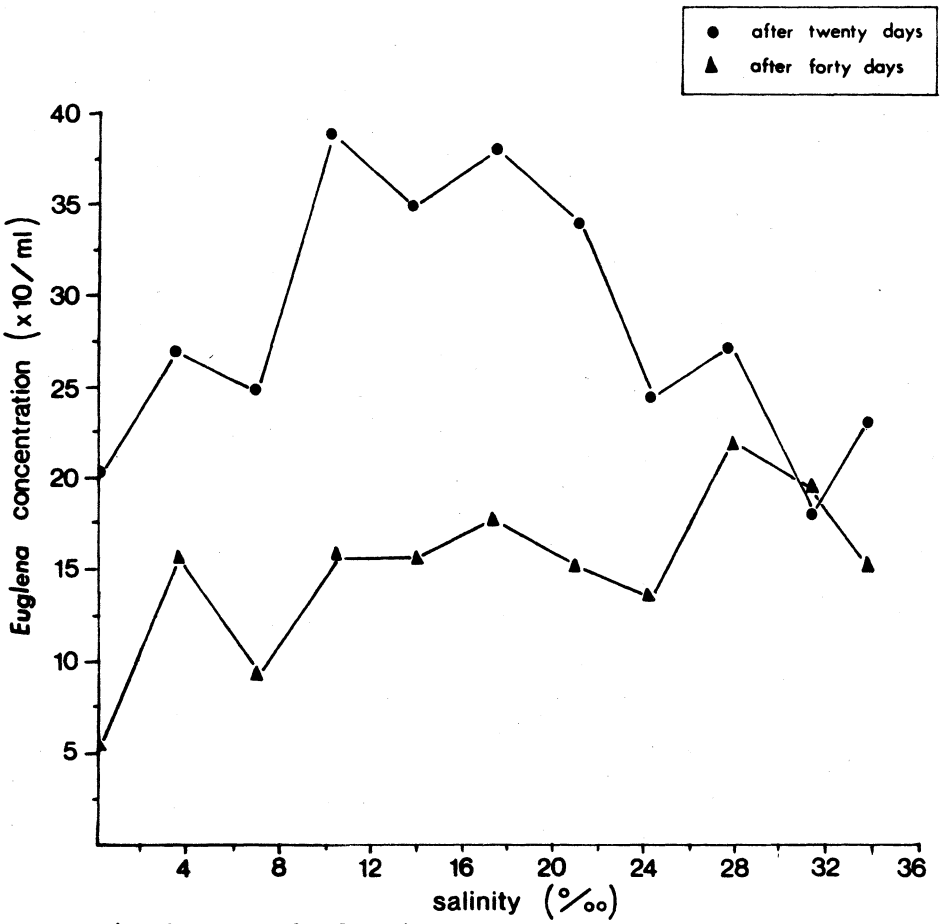

Fig. 9. Growth of *Euglena obtusa* in media of different salinities.

after two days in culture. Cameron (1970) thought that low salinity limited the upstream extension of *Euglena* in the Heathcote River. However, the results from this study indicate that although *E. obtusa* reached its highest density at intermediate salinities, it can survive in culture and in nature at levels very close to freshwater. The upstream limit of *E. obtusa* appears to be determined by the limits of the intertidal mud bank rather than by salinity.

The relationship between the water content of mud and *Euglena* numbers has been studied by other workers. Bracker (1929) found maximum numbers of *E. limosa* at 89% water content but only half the numbers at 82%. Bruce (1953) stated that maximum numbers occurred at 70-80% water content. These figures are 2-3 times higher than those found during the present study in which densities of 200 000 cells per 100 mm^2 were found on muds with only 14% water content. Water content therefore appears to be less important than previously assumed.

The factor most consistently linked with high *Euglena* numbers is high carbon content of the mud and in the Estuary most areas of high carbon concentration are found close to sewage effluent discharge points.

There are, however, a number of other factors which change along with carbon levels in the mud. Bruce (1953) noted that nitrogen levels in the mud followed the organic matter levels and were related to *Euglena* densities on the Estuary. Cameron (1970) found a similar relationship in the Lower Heathcote River. Phosphorus levels tend to follow the nitrogen and carbon levels (Knox and Kilner 1973). When the Estuary is compared with areas in the Lower Heathcote River, however, there is a considerable variation in nitrogen levels associated with very dense areas of *Euglena*. Cameron (1970) recorded 150 000 cells/100 mm^2 on muds containing 32 mg N/g. Bruce (1953) found a maximum of 100 000 cells/100 mm^2 on muds containing 750 mg N/g and very low concentrations of *Euglena* on muds with 280 mg N/g. Knox and Kilner (1973) quote levels of 250-500 mg N and over 500 mg P per g sediment in the *Euglena* areas on the Estuary while areas having low concentrations of *Euglena* had over 60 mg N and over 300 mg P per g. The interstitial water, which is the immediate environment of the *Euglena* has a more variable N level, but follows the same pattern as the substrate. Interstitial water in dense *Euglena* areas had nitrogen values ranging from 25 to 150 g/m^2 (Knox and Kilner 1973). It appears that most of the sediments in the Estuary have nutrient levels considerably higher than those associated with very dense *Euglena* populations on the Lower Heathcote River. Even these values are well in excess of normal plant requirements, and I, therefore, suggest that *Euglena* is not responding to high nutrient levels associated with carbon, but rather to the carbon itself. A possible explanation for this response to carbon is that *E. obtusa* feeds heterotrophically, thus utilising organic carbon in the mud directly. Heterotrophy, both obligate and facultative, has been recorded in a number of *Euglena* species (Leedale 1967), and Provasoli (1948) has described viable races of *Euglena* artificially rendered colourless by treatment with streptomycin. *E. obtusa* from the Estuary lost all colour within three weeks in a medium containing 0.6 mg streptomycin/litre, and these cultures remained colourless and at high densities for three months. This indicates an ability to use organic carbon sources in the absence of photosynthesis, which, if it occurs in nature, may explain its association with highly organic muds.

LITERATURE CITED

BRACHER, R. 1919. Observations on *Euglena dese*. *Annals of Botany 27*: 93-108.

———— 1929. The ecology of the Avon banks Bristol. *Journal of Ecology 17*: 36-81.

BRUCE, A. 1953. Report on a biological and chemical investigation of the waters in the estuary of the Avon and Heathcote Rivers. Unpublished report, Christchurch Drainage Board, New Zealand. 45 pp.

CAMERON, J. 1968. Report on a biological survey of the lower reaches of the Heathcote River. Unpublished report, Christchurch Drainage Board, New Zealand. 41 pp.

———— 1970. Biological aspects of pollution in the Heathcote River. *New Zealand Journal of Marine and Freshwater Research 4*: 431-444.

CARTER, N. 1933. Algal flora of two salt marshes. *Journal of Ecology 21*: 128-207.

GARD, M. 1919. Biologie d'une nouvelle espece d'Euglene (*Euglena limosa*). *Compte Rendus 169*.

GOJDICS, M. 1953. *The genus Euglena*. University of Wisconsin Press, Madison. 268 pp.

LEEDALE, G. 1967. *Euglenoid flagellates*. Prentice Hall, Englewood Cliffs, New Jersey. 242 pp.

LINZEY, J.T. 1944. A short study of the hydrography of the estuary of the Avon and Heathcote Rivers, near Christchurch. *Transactions of the Royal Society of New Zealand 73*: 365-376.

MARSHALL, J.W. 1971. A biological re-evaluation of the pollution in the lower Heathcote River, Christchurch, New Zealand. Unpublished M.Sc. project in Zoology. University of Canterbury, New Zealand. 24 pp.

METSON, A.J. 1956. Methods of chemical analysis for soil survey samples. *New Zealand Department of Scientific and Industrial Research - Soil Bureau Bulletin 12*:: 207 pp.

PALMER, J.D. and ROUND, F.E. 1965. Persistent vertical migration rhythms in benthic microflora. 1. The effect of light and temperature on rhythmic behaviour of *Euglena obtusa*. *Journal of the Marine Biology Association, United Kingdom 45*: 567-582.

PROVASOLI, L., HUNTER, S.H. and SCHATZ, A. 1948. Streptomycin-induced chlorophyll-less races of *Euglena*. *Proceedings of the Society for Experimental Biology and Medicine 69*: 279.

ROSENBERG, D.R. 1963. Report on a biological investigation of the estuary of the Avon and Heathcote Rivers. Unpublished report, Christchurch Drainage Board, New Zealand. 18 pp.

WEBB, B.F. 1965. Report on a biological investigation of the estuary of the Avon and Heathcote Rivers. Unpublished report, Christchurch Drainage Board, New Zealand. 27 pp.

WILLIAMS, M. 1960. Report on a biological investigation of the estuary of the Avon and Heathcote Rivers. Unpublished report, Christchurch Drainage Board, New Zealand. 18 pp.